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Feasibility Study of Passive Aerosol Sampler for Bio-agent Detection

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Feasibility Study of Passive Aerosol Sampler for Bio-agent Detection

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Abstract
We propose to establish the feasibility of a passive aerosol sampler for bio-agent collection through laboratory experiments and theoretical analysis. The passive sampler, unlike the typical active sampler, does not require pumps and complex fixtures, and thereby allows for large-scale field monitoring not possible with current active samplers. We plan to conduct experiments using model (both biological and non-biological) aerosols generated in an instrumented test chamber and compare the particles collected on various passive samplers to conventional filter samplers, commercial aerosol measuring instruments and to conventional surface swipes. Theoretical analysis will be used to design prototype passive samplers and to compare experimental results with theory. A successful feasibility study will be used to seek outside funding for applications that will greatly enhance current LLNL programs such as NARAC's atmospheric dispersal modeling, NAI's programs in bio-agent monitoring in public locations and fixed sampling stations, and EPD's environmental monitoring and decontamination research h. In addition, the feasibility study will position us favorably for responding to new calls for proposals by NIH and EPA for large scale environmental studies

Experimental Approach
The objective of the feasibility study is to characterize sampler properties and environmental conditions that determine passive collection of aerosols. The experimental approach for achieving this objective consists of experiments in a test chamber using a selection of different aerosols and a range of chamber conditions. Passive and active aerosol collection devices are exposed to the different aerosols and the collection efficiencies of the two compared as a measure of passive collection for the specific aerosol and conditions. Feasibility is assessed with the demonstration of reproducible collection efficiencies and the identification of sampler properties that could be modified to optimize those efficiencies.

Test chamber: The test chamber (Figure 1) is a 1.5 x 1.5 x 8 ft plexiglass sectional chamber equipped with various instruments for measuring chamber operating conditions and aerosol concentrations. Room air is drawn into the chamber through a HEPA filter by a 1 hp blower and exhausted to the building exterior through a second HEPA filter. An air flow rate of 10 ft/min is used to approximate indoor air flow. Access to the chamber is obtained through 5 doors along one side of the chamber. Sampling ports are located every 6 inches along the length of two chamber walls and every 2 inches along the height of the chamber wall 6 inches from the front and back of the chamber. These ports are used to verify air mixing and velocity instruments throughout the chamber and as access for sampling to the chamber interior.

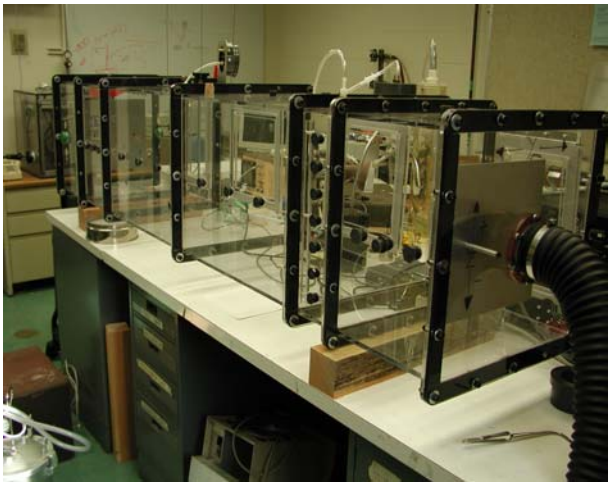


Figure 1: Test chamber used in the study. Room air and particles enter at the far end of chamber while the exhaust tube at the near end of chamber is connected to a HEPA filter. Black knobs along the vertical and horizontal axes of the chamber are sampling ports. Access doors are on the opposite chamber wall. Samples are fixed to scaffolding inside the chamber during experiments.

Model aerosols: Three types of aerosols are currently being tested in the chamber. Latex beads used as particles standards were obtained commercially. Fluorescein particles are generated using a standard

methodology. *Bacillus globigii* (BG) spores were obtained from Dugway Proving Grounds. Model aerosols vary from 0.5 to 5 microns in diameter. Aerosol charge is neutralized by passing the aerosol stream through a Kr85 non-fissionable radioactive Class 1 sealed source before it enters the chamber.

Aerosol Generation: Aerosols are generated in the chamber using two different generators. A nebulizer aspirates a liquid suspension of aerosols with compressed air flow. The latex beads and BG spores are aerosolized with this method. A vibrating orifice generator (VOG) is used to generate fluorescein aerosols. The VOG operates by forcing a fluorescein solution (water/isopropanol) through a 20 μm orifice that is intermittently passed through the stream. Uniform droplets are aspirated into a drying tube in which the solvent evaporates, leaving pure fluorescein particles. Setting VOG controls to manufacturer-specified values and alternating the fluorescein concentration can produce monodisperse particles ranging from 1 to 15 μm.

Aerosol Sampling and Analysis: Aerosols in the chamber are sampled by three methods. Real-time aerosol measurement is obtained with an aerodynamic particle sizer (APS 3000, TSI, Inc.) and a laser particle spectrometer (Particle Measuring Systems, Inc.) by drawing air samples through tubes that penetrate the chamber walls. Integrated air samples are collected by two methods. Active air sampling is performed during experiments by drawing air at various rates (2-15 L/min) through filter cassettes holding 37-mm, 0.4μm filters. Passive air sampling is conducted by affixing electrostatically charged polyethylene films perpendicular and parallel to the chamber airflow. After experiments with both sampling methods, aerosols are desorbed from the filters and films into an aqueous solution. The air samples are analyzed by two methods. Latex beads and BG spores are counted in a by a particle counter (Z-1, Coulter Counter). Flourescein particles are washed from the filters and films and quantified using a spectrofluorimeter and standard curves.

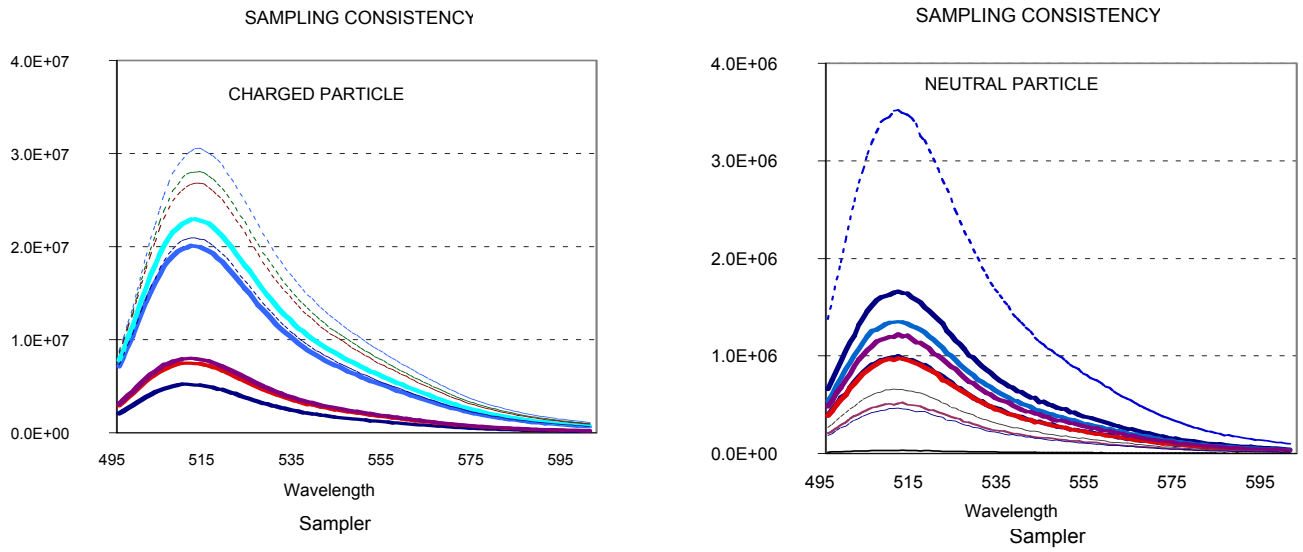
Scientific Progress

The scientific progress of the study has been delayed by programmatic and technical issues. Programmatic issues pertain to the safety review and procedures required for the research to be performed and center mainly on the aerosolization of biological materials in the B281 laboratory in which the study is performed. Hazards Control determined that the proposed research did not fall under the scope of work permitted by the 281 Facility Safety Plan and required that an Operation Safety Procedure (OSP) be prepared before the study commence. Considerable effort was required by the investigators to prepare the OSP. At the recommendation of Hazards Control, an application for review of the proposed research was submitted to the LLNL Institutional Biosafety Committee (IBC). BG is classified by CDC as a biosafety level 1 agent requiring minimal biosafety controls. However, the IBC determined that the aerosolization of biosafety level 1 agents elevates such agents to biosafety level 2 which require more stringent safety controls. Implementation of these safety controls required modifications to the laboratory and development of new experimental procedures. Technical difficulties arose with the VOG system and the BG spores. VOG manufacturer specifications did not produce monodisperse particles of the stated size and considerable effort was expended determining the instrument settings and operation required to produce particles in the size range required for the study. The size of the BG spores was more heterogeneous than expected and the development of additional procedures to prepare these particles for use was required. Although difficulties with the VOG system and BG spores limited the study of particle size as a parameter of collection efficiency, the collection of the bulk materials does allow a relative comparison of sampling efficiency.

Initial work has focused on determining the homogeniety of particle mixing within the chamber. Homogeniety is established with measurements of air velocity and particle concentration determined by the APS along x and y planes in the chamber. Modifications to the chamber were necessary based on this initial work. Experiments with the model aerosols are also being carried out. Examples of data obtained with the two analytical methods are shown in Figures 2 and 3. Due to the size heterogeniety of BG, experiments are conducted in which a solution containing BG and a latex bead standard (2.05 μm) is aerosolized in the chamber. The size of BG spores range from 0.7 - 1.5 μm whereas the size of the latex beads are exclusively in the 2 – 2.2 μm range. Particles are collected by the filter and film methods and two size ranges counted on the Coulter Counter.

Particles between 1 – 2.2 and 2 – 2.2 µm in diameter are counted and the ratio of the counts in 2 – 2.2 size range to counts in the 1 – 2.2 size range used as a measure of consistency within a sampling method and between the different sampling methods. The samplers did not consistently collect particles of the different size ranges, either within or between sampling methods (left). Modifications to improve air mixing within the chamber did not improve consistency (right).
Figure 2: Particle counts of film and filter samplers. Latex bead values increased by factor of 10 for scale.

Similar results are observed in experiments with the fluorescein particles (Figure 3). Particles generated by the VOG ranged from 1 to 5 µm so precise determination of the particle size collected by the two samplers is not possible. However, intra- and inter-sampler consistency can be assessed by the total fluorescein mass collected



on the filters and films as determined by the fluorescent intensity of the solutions used to extract the samplers. Consistent particle collection was not observed by either sampling method with charged or neutral particles.
Figure 3: Fluorescent intensity of film and filter samplers after fluorescein collection. Thick lines represent filter samples and thin lines represent film samples.

Conclusion

The assessment of the feasibility of passive samplers for bio-agent collection conducted by this study was marginally successful. The study did demonstrate that BG spores, a widely used bio-agent simulant, are collected quite effectively on passive. Large particle counts were observed on the films and filters following BG aerosolization. However, sample collection was not consistent nor in agreement with expected results. Passive collection equaled and exceeded active collection in most experiments. The study was hampered by technical deficiencies and efforts are underway to correct these deficiencies. The VOG instrument has been modified and calibrated to produce consistent aerosol size and concentration. Methods to separate the BG spores into more discreet size fractions are being developed. Additional studies of chamber mixing are being conducted. For this study, the chamber was operated at a very low air speed and higher air speeds will be used to simulate passive collection of outdoor air conditions. Better methods for particle identification are being developed. Particle counting in the size range used in this study are at the limits of the instrument and highly influenced by background. Additional studies with BG and other aerosols are planned.

Exit Plan

This study will support existing LLNL programs in NAI for bio-detection, in NARAC for validating dispersal and transport codes, and in EPD for ambient monitoring in buildings and outdoors, and Energy & Environment for particle sampling related to dose reconstruction. The B281 particle laboratory is one of the few laboratories

on site with IBC approval to aerosolize BG and as such it will be an important resource to other LLNL investigators. The investigators are in discussions with other groups at LLNL to use the particle laboratory for their research projects, in particular for additional testing of the sampling train of the Autonomous Pathogen Detection System and for evaluation of decontamination protocols developed by EPD. In addition, the investigators have been asked to evaluate passive sampling devices for an asthma study being conducted by the School of Public Health, University of California at Berkeley. The UC investigators have also expressed an interest in developing low-cost, passive dust sampling devices for a multi-center, national asthma study called the Children's Health Study. This NIH-supported study is projected to last 20 years and is designed to collect dust samples from 100,000 homes.